# Evidence for biological nature of the grape replant problem in California

Andreas Westphal<sup>1,4</sup>, Greg T. Browne<sup>2</sup> & Sally Schneider<sup>3</sup>

<sup>1</sup>Department of Plant Pathology, University of California, Davis. <sup>2</sup>USDA-ARS, Davis, California, USA. <sup>3</sup>USDA, Fresno, California, USA. <sup>4</sup>Corresponding author

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#### **Abstract**

A bioassay was developed to investigate causes of grape replant problems under controlled conditions. Soils were collected from methyl bromide-fumigated and non-fumigated plots at a site cleared from a 65-year-old grape vineyard (Vitis vinifera cv. Thompson seedless) at Parlier, CA. Subsamples of the non-fumigated soil were either left non-treated, subjected to autoclaving (twice 45 min), or heating at 40, 50, 60, 70, 80 or 90 °C for 30 min. Subsequently, the samples were placed in 120-mL pots, planted with rooted hardwood grape cuttings (V. vinifera, cv. Carignane) and placed in a greenhouse or growth chamber. Three months after transplanting, vines from nontreated or 40 °C-treated soil had lower shoot weights and densities of healthy lateral roots than vines from the other treatments. Pythium spp. were isolated from 45 to 55% of the plated root segments from vines grown in non-treated, or soil that had been heated at 40 or 50 °C but were not detected in roots from soil given other treatments. Egg masses of root-knot nematode, *Meloidogyne* spp., were produced on roots from non-treated or heated at 40 °C soil, but no egg masses were detected on roots of the other treatments. In another test with the same soils, remnant roots from non-fumigated or pre-plant methyl bromide-fumigated soil were extracted and amended to non-fumigated soil, soil from fumigated field plots, soil fumigated in a small container, or autoclaved potting mix. The transfer of old vine roots from non-fumigated field soil resulted in incidence of Pythium spp. on grape assay roots, but there was no measurable effect of the transfer on growth and health of the bioassay plant roots. The results of the bioassays indicate that grape replant problem at the California site had biological causes. The bioassay approach may aid in future determinations of the etiology of grape replant problems.

#### Introduction

Fruit production is located within limited geographic areas due to requirements of the crop for climatic and environmental conditions. Farms are often specialized on a limited number of fruit crops due to the economic design of the operations. This often results in repeated planting cycles of closely related tree or vine crops at individual orchard or vineyard sites. In many cases such replanting of apple, grape, peach and other crops has resulted in unthrifty and slow growing young plants (Deal et al., 1972; Hine, 1961; Mazzola, 1998; Traquair, 1984). Young grape plantings are affected by the replant problem; symptoms of which include poor and uneven top growth, and root browning (Brinker and Creasy, 1988).

The cause of replant problems is complex. Detailed studies of apple replant problems in Australia and the United States demonstrated the effect of several soil-borne fungal pathogens and the lesion nematode (Pratylenchus penetrans (Cobb) Chitwood and Oteifa) in the development of the apple replant disorder (Dullahide et al., 1994; Jaffee et al., 1982a; Mazzola, 1998). Other studies indicated the involvement of actinomycetes in the apple replant problem (Otto, 1973, Westcott et al., 1987). In the grape replant problem, causal roles have been suggested for hyphomyceteous fungi (e.g., Penicillium Link:Fr, Fusarium Link:Fr., Gliocladium Corda, Roesleria hypogaea Thuem. & Pass.) or fluorescent pseudomonads and the reduced infection with endomycorrhizal fungi (Deal et al., 1972; Waschkies et al., 1994). In addition, oomycetes, such as *Phytophthora* de Bary and *Pythium* Pringshein species can damage root systems of young grape plants (Chiarappa, 1959; Marais, 1979, 1980). The role of old grape roots from the previous crop in the vineyard soil is not clear as of yet; a negative effect of autotoxicity conferred by old grape roots was found in one study (Brinker and Creasy, 1988) while other investigations found positive effects of old roots in increased endomycorrhizal colonization and increased populations of other beneficial microorganisms (Deal et al., 1972). Plant-parasitic nematodes are also often associated with poor growth of young grape plants (McKenry et al., 1994). Fine-textured soils and soils with higher organic matter contents were less conducive to the replant problem (Moser, 1963).

Although fruit crop replant problems are often reduced by soil fumigation with methyl bromide (Mai and Abawi, 1981), this fumigant will be phased out and has not been available for soil fumigation in Europe for the last decade. One of the alternative fumigants, methyl iodide has similar efficacy as methyl bromide in reducing replant problems in peach (Eayre et al., 2000). Currently, registration for crop protection is sought. Other compounds are currently being tested under efforts to identify alternatives to methyl bromide, however, a comprehensive understanding of the etiology of the grape replant problem is still lacking. To facilitate the development of new control methods, we need a better understanding of what causes the problem.

The current project evaluated the effects of manipulation of the microbiota of old vineyard soil on grape assay plants in controlled and reproducible tests, which may constitute a bioassay technique for evaluating replant problems. The application of heat, which selectively reduced microorganisms with varied temperature sensitivity (Baker and Roistacher, 1957), was chosen to accomplish these manipulations. This method was used because of the demonstrated potential in studies of other complex soil microbial communities (Rouxel et al., 1977; Westphal and Becker, 2001). Additionally, a root transfer test was included in an attempt to clarify the role of old grape roots in transmission of the disease complex. A preliminary report of this study has been published (Westphal et al., 2000).

#### Materials and methods

Soil source

Soil samples were collected from a field trial arranged in a randomized complete block design from each of five replicates of non-fumigated or methyl bromidefumigated (450 kg ha<sup>-1</sup>) plots at a 65-year-old vineyard site (Vitis vinifera L. cv. Thompson seedless) at the USDA-ARS research station in Parlier, CA. Soil fumigation was conducted by a commercial applicator utilizing tractor-pulled application shanks spaced 150-cm apart, inserted 50-cm in the ground during the application of commercial methyl bromide (995 g kg<sup>-1</sup> methyl bromide, 5 g kg<sup>-1</sup> chloropicrin) and covering the soil with 0.3-mm polyethylene tarp. The soil was a Hanford fine sandy loam soil. The vineyard had been removed in September 1999, soil fumigation was applied on 13 November 1999, and soil samples were taken in January 2000. Collections were made from a 0 to 45 cm depth, placed in plastic containers, transported to Davis, CA and stored at  $5 \pm 2$  °C until used for greenhouse and growth chamber tests. Cold storage of the soils lasted between 1 to  $2\frac{1}{2}$  months.

# Planting material

Grape planting material was collected and prepared according to standard procedures as utilized by the Department of Viticulture, UC Davis (Richard Hoenisch, personal communication). One-year-old shoots of grape (Vitis vinifera cv. Carrignane) were collected in January 1999. After 12 h soaking in water, the scions were dipped for 5 min in 101 g kg<sup>-1</sup> commercial bleach solution (52.5 g kg<sup>-1</sup> NaOCl), rinsed with water and dipped in lime sulfur solution (20 g kg<sup>-1</sup> calcium sulfide). The cuttings were placed in plastic bags and stored at 1°C until use. Two weeks before planting, scions were soaked in water for 12 h, and cut to 2-bud length. The scions were imbedded in a wellmoistened peat moss-perlite mixture (50% + 50%) and incubated at 27 °C. After three weeks, the cuttings had developed callus tissue and adventitious roots. The upper end of the scion was covered with wax (67 °C melting point). The rooted 2-bud cuttings were planted either directly into the experimental soils or planted into potting mix (Redi-Gro Corporation, Sacramento, CA, 0.4 m<sup>3</sup> no. 2 washed sand, 0.2 m<sup>3</sup> nitrified redwood compost, 0.2 m<sup>3</sup> sphagnum peat moss, 0.2 m<sup>3</sup> pumice rock,  $4.15 \text{ kg m}^{-3}$ dolomite lime,  $1.48 \text{ kg m}^{-3}$ oystershell lime, 1.48 kg m<sup>-3</sup> superphosphate, 0.59 kg m<sup>-3</sup> calcium citrate, 0.22 kg m<sup>-3</sup> potassium nitrate, and  $0.15 \text{ kg m}^{-3}$  potassium sulfate) that were later planted into the experimental soils.

#### Heat treatments of old vineyard soil

Soils from both non-fumigated and methyl bromidefumigated areas were sieved (3-mm openings). Root pieces retained on the screen were cut into  $\leq 1$ -cm pieces, and mixed back into the soil of origin. Samples of the non-fumigated soil were subdivided into 150mL portions, and were placed into double envelopes of two polyethylene bags. Ten such non-fumigated soil portions were either not treated or were heated in a waterbath (30 L-capacity) at 40, 50, 60, 70, 80 or 90 °C. Increase of the soil temperature was monitored with a laboratory thermometer which was inserted into one of the soil samples. When the desired treatment temperature was reached (after ca. 12 min), soil samples were kept for 30 min at that temperature in the waterbath. Another portion of the non-fumigated soil was autoclaved for 45 min on two consecutive days. Similar samples were taken from the methyl bromidefumigated soil. The methyl bromide-fumigated soil was not heated.

After treating, soil samples were stored at  $5 \pm 2$  °C for  $\leq 2$  days and subsamples of the soil (n = 5) were dilution plated on modified selective media for fluorescent pseudomonads (Sands and Rovira, 1970; 10 mL glycerol, 1.5 g K<sub>2</sub>HPO<sub>4</sub>anhydrous, 1.5 g MgSO<sub>4</sub> · 7 H<sub>2</sub>O, 10 g proteose peptone No. 3, 20 g agar in 1 L water, and 45 mg novobiocin and 45 mg penicillin g were diluted in methanol and added after autoclaving when media was cooled to 45–48 °C). Colonies of fluorescent pseudomonads were counted under long wave-length UV light after 48-72 h incubation at room temperature. At the same time, the bulk of the samples were placed into 120-mL polyethylene cones (3.8 cm in diameter, 20.6 cm deep, Stuewe and Sons Inc., Corvallis, OR). Each cone was planted with a freshly rooted hardwood cutting of grape, and treated soil was covered with a 5-cm layer of potting-mix as a barrier to reduce splashing during watering. Treatments were arranged in a randomized complete block design with 10 replications. The first test was placed onto propagation-mats (Pro-Grow Supply Company, Brookfield, WI) to reduce temperature variation in a heated greenhouse (average: 23 °C, range: 17–38 °C). The second test was placed into a growth chamber with 16/8 h day/night cycle at 24/18 °C and incandescent light (530  $\mu$ mol). During the growing period plants were fertilized weekly with

10 to 20 mL nutrient solution (Miracle Gro, 2.64 g  $L^{-1}$  of water; 150 g kg<sup>-1</sup> N, 130 g kg<sup>-1</sup> P, 120 g kg<sup>-1</sup> K, and micronutrients, Scotts Miracle Gro Products, Port Washington, NY). After three months, the newly grown grape shoots were cut off and weighed, and the root systems were carefully removed from the cones and carefully rinsed under running tap water. The root systems were visually inspected for replant problem symptoms, and root-knot nematode eggmasses were counted on the entire root systems after staining in erioglaucine solution (133 mg L<sup>-1</sup>, Sigma St. Louis, MO) (Omwega et al., 1988). For each root system, five adventitious roots were chosen at random, numbers of fine roots per 5-cm root length (5-10 cm depth) were determined, and the physiological status of the fine roots was classified as healthy (yellow-brown) or diseased (dark-black and necrotic). Single nematode females were picked from the roots and identified to species utilizing the phastgel procedure (Esbenshade and Triantaphyllou, 1985). Twenty random 1-cm root pieces of each of three random replicates were excised from the root system (5-cm depth), dipped into ethanol solution (700 g L<sup>-1</sup> ethanol in water) for 30 s and imbedded into modified PARP media (Browne and Viveros, 1999), which is semi-selective for oomycetes. Emerging Pythium colonies were counted and expressed as percent of root pieces with infection.

## Transfer test via old roots from grape replant vineyard

Soils from the non-fumigated and methyl bromidefumigated plots near Parlier, CA were also used for a root transfer study. In this test, root pieces were only mixed back into the following treatments: fieldfumigated vineyard soil, non-treated vineyard soil, potting mix, and originally non-treated vineyard soil that had been fumigated in small containers (400 kg ha<sup>-1</sup>) (Becker et al., 1998). Additional non-amended treatments served as controls (Table 2). Soils were then placed into 500-mL polyethylene pots and arranged in a randomized complete block design with eight replications in the greenhouse using conditions similar to those of the first greenhouse test. The sum of weights from prunings above the second basal bud during the season (4 and 8 weeks after planting), and the shoots at harvest (16 weeks after planting) were reported as accumulated shoot dry weight. The prunings were done to reinvigorate growth. Plants were fertilized weekly with 40 mL of a nutrient solution (Miracle Gro,  $2.64 \text{ g L}^{-1}$  of water). At harvest, roots were washed free of soil, weighed, and feeder root health

for ten randomly chosen 5-cm root sections from below potting soil cover was determined as described for the first test. The root systems were also inspected for possible nematode symptoms. Ten randomly chosen 1-cm root sections were plated on PARP for the isolation of oomycetes as described for the heat treatment experiments.

### Data analysis

Data were analyzed utilizing GLM (SAS Institute, Cary, NC) followed by Waller-Duncan mean separation at P = 0.05. For the heat treatment experiments, the interaction experiment  $\times$  replication was found non-significant and the two tests were calculated combined. There were significant interactions of treatment  $\times$  experiment among the heat treatment experiments. However, the statistical groupings within each experiment calculated separately did not differ in their biological implications.

#### Results

Heat treatments of old vineyard soil

Heat treatments reduced fluorescent pseudomonad populations at temperatures  $\geq 50$  °C. The non-treated and the 40 °C treatments had lower accumulated shoot dry weights than any other treatment (Table 1). The root fresh weights were higher in the non-treated, field-fumigated, 40 and 50 °C treatments than in the 90 °C and the autoclaved treatment. The number of rootlets cm<sup>-1</sup> root was the lowest in the 40 °C and the non-treated control, and somewhat elevated in the 50 °C treatment. But the percentage of diseased fine roots of the total number of fine roots was the greatest in the non-treated and 40 °C treatments, and was elevated in the 50 °C treatment in comparison to heat treatments  $\geq$  60 °C (Table 1). The largest numbers of egg masses of *Meloidogyne* spp. were in the 40 °C and non-treated control. Meloidogyne javanica (Treub) Chitwood was identified utilizing the phast gel technique. A second root knot nematode was found but not identified. Pythium spp. were present on roots from the non-treated, 40 and 50 °C treatments and almost absent in the other treatments.

Transfer test via old roots from replant problem vineyard

Cuttings grown in the non-treated old vineyard soil (soil and roots) had lower accumulated shoot dry

weights than the other treatments with exception of the old vineyard soil from which root pieces were removed (Table 2). Root fresh weights were similar in all field soils and were higher in potting-mix treatments than in the field soils. The number of rootlets cm<sup>-1</sup> root was smaller in the replant problem soil than in any other treatments and was higher in the potting-mix treatments than the field soils. The ratio of diseased to healthy rootlets cm<sup>-1</sup> root was larger in the replant problem soil than in other treatments. Incidence of *Pythium* spp. on assay roots was higher in replant problem soil with or without old roots and in field-fumigated soil amended with roots derived from replant problem soil than in the other treatments. No *Meloidogyne* spp. was detected.

## Discussion

Symptoms resembling those described for fruit replant problems (Mai and Abawi, 1981) were observed in small pots when grape cuttings were planted into old vineyard soil. Fine roots of diseased plants were fewer in number and were discolored and/or necrotic in contrast to roots in fumigated or heat-treated soil. The elimination of lateral root necrosis at low temperatures suggested the involvement of soil microbial groups in the development of the disease symptoms and was not consistent with the involvement of autotoxic compounds as suggested for the grape replant problems in New York state (Brinker and Creasy, 1988).

Higher incidence of recovery of *Pythium* spp. from roots was observed when old root pieces from replant problem soil were transferred into the test soil, but this increased incidence in recovery did not always result in feeder root necrosis. Different species of Pythium were considered important components of the complexes causing replant problems in apple or peach. In Washington State, P. sylvaticum Campbell and Hendrix and P. ultimum Trow were isolated from replant problem soil-grown apple roots (Mazzola, 1999). Pythium irregulare Buisman was frequently found on apple roots grown in replant soil in State New York (Jaffee et al., 1982b). In Australia, one unidentified species of Pythium was isolated most frequently from apple roots (Dullahide et al., 1994). Pythium ultimum was found associated with the peach replant problem in California (Hine, 1961). Oomycetes were important in root rot and decline of grape in California (Bayramian et al., 1998; Chiarappa, 1959) and South Africa (Marais, 1979, 1980) but when indicated in re-

Table 1. Effect of pre-plant fumigation or various heat treatments on certain microorganisms and the growth of grape hardwood cuttings (Vitis vinefera cv. Carrignane) in soil collected from a vineyard replant site<sup>a</sup>

Treatment	Fluorescent Pseudomonads <sup>b</sup> (cfu g 10 <sup>-5</sup> )	Accumulated shoot dry weight <sup>c</sup> (g)	Root fresh weight <sup>c</sup>	Number of rootlets per cm <sup>cd</sup>	Incidence of diseased fine roots (%) <sup>ce</sup>	RKN egg masses per root system <sup>f</sup>	Pythium (%) <sup>g</sup>
Non-treatedhh	6.8 b	3.1 d	7.8 a	1.78 d	41.9 a	85.9 a	55 a
Field-fumigated <sup>i</sup>	12.7 a	3.6 ab	7.5 ab	2.33 bc	0.4 b	1.5 b	0 b
$40  {}^{\circ}\text{C}^{h}$	6.3 b	3.1 cd	7.5 ab	1.80 d	41.9 a	91.5 a	53 a
50 °C <sup>h</sup>	0.6 c	3.8 a	7.8 a	2.10 cd	8.5 b	5.3 b	45 a
60 °C <sup>h</sup>	0.1 c	3.5 b	6.6 bc	2.59 ab	0.6 c	0.0 b	0 b
70 °C <sup>h</sup>	0.0 c	3.6 ab	6.9 abc	2.84 a	0.3 c	0.0 b	0 b
$80  {}^{\circ}\mathrm{C}^{h}$	0.0 c	3.5 b	6.8 abc	2.42 bc	0.8 c	0.0 b	0 b
90 °C <sup>h</sup>	0.0 c	3.5 b	6.2 c	2.49 ab	0.6 c	0.0 b	0 b
Autoclaved <sup>h</sup>	0.0 c	3.4 bc	6.1 c	2.44 bc	0.2 c	0.0 b	1 b
MSD for $P = 0.05$	1.1	0.3	1.2	0.36	6.4	19.6	20
P for treatment $F$	0.0001	0.0001	0.0035	0.0001	0.0001	0.0001	0.0001

<sup>&</sup>lt;sup>a</sup> Data of one greenhouse and one growth chamber experiment which each had 10 replications were combined. Water bath heat treatments were applied for 30 min. Treatments followed by the same letter within a column were not significantly different when tested with Waller at P = 0.05. The minimum significant difference (MSD) is given for this comparison.

plant problems occurred together with other soil-borne pathogens (Dullahide et al., 1994; Hine, 1961; Jaffee et al., 1982b; Mazzola, 1999). Other components of the soil-borne complex were possibly not transferred with the old vineyard roots, but would have been instrumental for replant problem to occur.

Root-knot nematodes, which were found in the heat treatment experiments after ≤50 °C, have been observed associated with California grape production, particularly in coarse-textured soils (Ferris and McKenry, 1975). Major efforts in resistance breeding towards *Meloidogyne incognita* are underway(Walker et al., 1994). *Meloidogyne incognita* had been considered as one problem in establishing new grape plantings in old vineyard sites and its presence was the reason for utilizing 1,3-D and methyl bromide treatments for improving such plantings (Raski et al., 1973). However, in this investigation *Meloidogyne* spp. were not consistently associated with lateral root necrosis; symptoms occurred whether or not root-knot nematodes were detected.

Heat treatments of  $\geq 50$  °C improved health of the fine roots. Selectivity of heat treatments at 40 and 50 °C ranges was demonstrated by quantifying the colony forming units of fluorescent pseudomonads. The improvement of root health occurred with considerably less heat than necessary to reduce replant problem of apple or other replant problems (60 °C for 1 h, Otto, 1972b). Selective heat kill of microorganisms has been used in surveys, soil pasteurization at 70 °C for 1 h was used to survey fields for the presence of replant problems (Utkhede and Thomas, 1988). The difference in treatment time might have been due to the fact that we measured the time of actual treatment temperature reached within the soil for duration, while Otto (1972b) only measured the exposure time to the temperature. In addition, different organisms may be involved in the replant problem of apple compared to the grape replant problem.

Utilization of old grape roots collected from the bulk soil simulated the field situation of complete destruction of the old vine crop and replanting into soil containing residual roots. Amendment of non-replant

<sup>&</sup>lt;sup>b</sup> Following heat treatments, soil samples were dilution plated on NPC media and fluorescent colony forming units (cfu) of Pseudomonads were counted under long wave-length UV light after 48-72 h, n=5 per each of two experiments.

 $<sup>^{</sup>c}$  n = 10 per each of two experiments.

 $<sup>^{</sup>d}$  Total number of fine roots on five randomly chosen 5-cm adventitious roots per root system.

<sup>&</sup>lt;sup>e</sup> Percentage of fine roots that were either dark-black or necrotic of the total number of fine roots.

f n = 5 per each of two experiments.

g Percentage of 20 random root pieces that yielded *Pythium* spp. when plated on PARP media, n = 3 per each of two experiments.

<sup>&</sup>lt;sup>h</sup> Non-treated soil from 65-year old vineyard 4 months after removal of a planting of Thompson seedless grape, either non-treated, heated at the temperature indicated for 30 min or autoclaved twice (45 min each, with 24 h between heatings, 121 °C).

<sup>&</sup>lt;sup>i</sup> Soil from the same vineyard but fumigated with methyl bromide in the field (450 kg ha<sup>-1</sup>).

Table 2. Growth of grape hardwood cuttings (Vitis sp.) in soil collected from an old vineyard replant site after different fumigation treatments and root transfers from non-treated and pre-plant fumigated soil<sup>a</sup>

Soil $treatment^b$	Root treatment <sup>c</sup>	Accumulated shoot dry weight $^d$ (g)	Root fresh weight $^d$ (g)	Number of fine roots per $cm^{de}$	Incidence of diseased rootlets $(\%)^{df}$	Pythium (%) <sup>g</sup>
RPP	+	6.7 d	18.3 c	1.84 d	20.9 b	95 a
RPP	-	7.8 cd	18.2 c	1.80 d	26.2 a	79 a
Ffum	+	10.0 ab	19.6 c	2.64 c	1.0 c	17 b
Ffum	-	9.0 bc	17.6 c	2.72 c	0.6 c	25 b
Bfum	+	9.2 b	19.4 c	2.43 c	1.3 c	0 b
Bfum	-	9.2 b	17.4 c	2.47 c	0.1 c	5 b
Ffum	+from RPP	10.0 ab	17.5 c	2.30 c	4.1 c	80 a
Pot-mix	+from RPP	10.1 ab	23.0 b	3.74 ab	1.7 c	33 b
Pot-mix	-	10.1 ab	23.8 ab	3.81 a	1.5 c	0 b
Pot-mix	+from Ffum	10.6 a	25.4 a	3.38 b	1.2 c	3 b
MSD for $P = 0.05$		1.2	2.4	0.42	4.1	37
P for treatment $F$ 0.0001		0.0001	0.0001	0.0001	0.0001	

<sup>&</sup>lt;sup>a</sup> Data of one greenhouse experiment, treatments followed by the same letter within a column were not significantly different when tested with Waller at P = 0.05. The minimum significant difference (MSD) is given for this comparison.

problem soil with roots from replant problem soil has several difficulties because the distinction between the transfer of toxic break-down products and microbial communities is difficult (Otto, 1972a). In our test, removal of roots from replant problem soil did not result in increased numbers of rootlets and more of the assay rootlets were diseased. In apple replant problem, the transfer of live root systems was implemented and transfer of replant problem was observed (Otto, 1972a). We did not make this observation, but data suggest that old grape roots in the development of replant problem symptoms were less important in this soil. In particular, the number of fine roots and fine root health did not decrease when old grape roots were added to non-replant soil.

The possibility to study grape replant problem in small containers will be critical to studying larger numbers of soil treatments under contained and controlled conditions. Small container tests have been successfully used in advisory systems for the management of the replant problem of apple. In these systems, orchard soils are collected and tested in the greenhouse for replant problem and different soil amendments are tested for their potential to reduce the problem (Neilsen et al., 1991; Slykuis, 1990). Tests in small

containers have a potential benefit in mitigating losses due to grape replant problem by developing an advisory system for grape replant problem management. In addition, such test should be useful for studies of the etiology of this complex soil-borne problem.

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## References

Baker K F and Roistacher C N 1957 Heat treatment of soil. *In* The UC system for producing healthy container-grown plants. Ed. K F Baker. pp 123–137. University of California, Division of Agricultural Sciences, Oakland, CA.

<sup>&</sup>lt;sup>b</sup> Soil was either non-treated vineyard soil (RPP); methyl bromide-fumigated vineyard soil (450 kg ha<sup>-1</sup>) (Ffum); methyl bromide-fumigated in a small container (390 kg ha<sup>-1</sup>) (Bfum) or Pot-mix (40% sand, 20% compost, 20% peat moss, 20% pumice rock and plant nutrients).

<sup>&</sup>lt;sup>c</sup> Roots were derived from RPP or Ffum soil and were mixed into the receiving soil at the rate they had been recovered from the source soil,

<sup>+:</sup> roots amended; -: roots not amended.

 $<sup>^{</sup>d}$  n = 8.

 $<sup>^{\</sup>it e}$  Fine roots counted on 10 randomly chosen 5-cm adeventitious roots per root system.

f Percentage of fine roots that were either dark-black or necrotic of the total number of fine roots.

<sup>&</sup>lt;sup>g</sup> Percentage of 10 random root pieces that yielded *Pythium* spp. when plated on PARP media, n = 4.

- Bayramian L A, Browne G T and Walker M A 1998 *Phytophthora* and *Pythium* spp. as pathogens of grape in California. (Abstract) Phytopathology 88, S6.
- Becker J O, Ohr H D, Grech N M, McGiffen M E and Sims J J 1998 Evaluation of methyl iodide as a soil fumigant in container and small field plot studies. Pestic. Sci. 52, 58–62.
- Brinker A M and Creasy L L 1988 Inhibitors as a possible basis for grape replant problem. J. Amer. Soc. Hort. Science 113, 304– 309.
- Browne G T and Viveros M A 1999 Lethal cankers caused by *Phytophthora* spp. in almond scions: specific etiology and potential inoculum sources. Plant Dis. 83, 739–745.
- Chiarappa L 1959 The root rot complex of Vitis vinifera in California. Phytopathology 49, 670–674.
- Deal D R, Mai W F and Boothroyd C W 1972 A survey of biotic relationships in grape replant situation. Phytopathology 62, 503– 507.
- Dullahide S R, Stirling G R, Nikulin A and Stirling A M 1994. The role of nematodes, fungi, bacteria, and abiotic factors in the etiology of apple replant problems in the Granite Belt of Queensland. Aust. J. Exp. Agric. 34, 1177–1182.
- Eayre C G, Sims J J, Ohr H D and Mackey B 2000 Evaluation of methyl iodide for control of peach replant disorder. Plant Dis. 84, 1177–1179.
- Esbenshade P R and Triantaphyllou A C 1985 Use of enzyme phenotypes for identification of *Meloidogyne* species. J. Nematol. 17, 6–20
- Ferris H and McKenry M V 1975 Relationship of grapevine yield and growth to nematode densities. J. Nematol. 7, 295–304.
- Hine R B 1961 The role of fungi in the peach replant problem. Plant Dis. 45, 462–465.
- Jaffee B A, Abawi G S and Mai W F 1982a Role of soil microflora and *Pratylenchus penetrans* in an apple replant disease. Phytopathology 72, 247–251.
- Jaffee B A, Abawi G S and Mai W F 1982b Fungi associated with roots of apple seedlings grown in soil from an apple replant site. Plant Dis. 66, 942–944.
- Mai W F and Abawi G S 1981 Controlling replant diseases of pome and stone fruits in Northeastern United States by preplant fumigation. Plant Dis. 65, 859–864.
- Marais P G 1979 Fungi associated with root rot in vineyards in the western cape. Phytophylactica 11, 65–68.
- Marais P G 1980 Fungi associated with decline and death of nursery grapevines in the western cape. Phytophylactica 12, 9–13.
- Mazzola M 1998 Elucidation of the microbial complex having a causal role in the development of apple replant disease in Washington. Phytopathology 88, 930–938.
- McKenry M, Buzo T, Kretsch J, Kaku S, Otomo E, Ashcroft R, Lange A and Kelly A 1994 Soil fumigants provide multiple benefits; alternatives give mixed results. California Agriculture 48, 22–28
- Moser L 1963 Versuche zur Bekämpfung der Rebmüdigkeit. Klosterneuburg Mitteilungen Ser. A 13.
- Neilsen G H, Beulah J, Hogue E J and Utkhede R S 1991 Use of greenhouse seedling bioassays to predict first year growth of apple trees planted in old orchard soil. HortScience 26, 1383–1386.

- Omwega C O, Thomason I J and Roberts P A 1988 A non-destructive technique for screening bean germplasm for resistance to *Meloidogyne incognita*. Plant Dis. 72, 970–972.
- Otto G 1972a Untersuchungen über die Ursache der Bodenmüdigkeit bei Obstgehölzen: I. Versuche zur Übertragung der Bodenmüdigkeit durch Wurzeln. Zentralblatt für Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene II 127, 279–289.
- Otto G 1972b Untersuchungen über die Ursache der Bodenmüdigkeit bei Obstgehölzen: III. Versuche zur Beseitigung der Bodenmüdigkeit durch Dämpfung bei verschiedenen Temperaturen. Zentralblatt für Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene II 127, 777–782.
- Otto G 1973 Untersuchungen über die Ursache der Bodenmüdigkeit bei Obstgehölzen: V. Einfluß unterschiedlicher Dämpftemperaturen auf die Mikroflora eines müden Bodens. Zentralblatt für Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene II 128:377–382.
- Raski D J, Schmitt R V, Luvisi D A and Kissler J J 1973 1,3-D and methyl bromide for control of root-knot and other nematodes in vineyard replants. Plant Dis. Rep. 57, 619–623.
- Rouxel F, Alabouvette C and Louvet J 1977 Recherches sur la résistance des sols aux maladies II. Incidence de traitements thermique sur la résistance microbiologique d'un sol à la Fusariose vasculaire du melon. Ann. Phytopathol. 9, 183–192.
- Sands D C and Rovira A D 1970 Isolation of fluorescent pseudomonads with selective medium. Appl. Microbiol. 20, 513–514.
- Slykuis J T 1990 Greenhouse testing of soils for treatments to assure good growth of young apple trees. Compact Fruit Tree 23, 145– 146.
- Traquair J A 1984 Etiology and control of orchard replant problems. Can. J. Plant Pathol. 6, 54–62.
- Utkhede R S and T S C Li 1988 Determining the occurrence of replant disease in British Columbia orchard and vineyard soils by pasteurization. Can. Plant Dis. Surv. 68, 149–151.
- Walker A, Ferris H and Eyre M 1994 Resistance in Vitis and Muscadinia species to Meloidogyne incognita. Plant Dis. 78, 1055–1058.
- Waschkies C, Schropp A and Marschner H 1994 Relations between grapevine replant disease and root colonization of grapevine (Vitis sp.) by fluorescent pseudomonads and endomycorrhizal fungi. Plant Soil 162, 219–227.
- Westcott III S W, Beer S V and Israel H W 1987 Interaction between actinomycete-like organisms and young apple roots grown in soil conducive to apple replant disease. Phytopathology 77, 1071–1077
- Westphal A and Becker J O 2001 Components of soil suppressiveness against *Heterodera schachtii*. Soil Biol. Biochem. 33, 9–16.
- Westphal A, Browne G T and Schneider S 2000 Development of a bioassay to characterize grape replant problem. Phytopathology 90, S83

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